THE PATHOLOGICAL EFFECTS OF DEAD TUBERCLE BACILLI.

By STEWART STOCKMAN,

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EXPERIMENTS WITH DEAD TUBERCLE BACILLI.

Such experiments have been undertaken before. Koch [illegible] that dead tubercle bacilli produced an abscess in [illegible] when injected subcutaneously.

Prudden and Hodenpyl[1] experimented more fully on guinea-pigs and rabbits. In their experiments they used dead tubercle bacilli from cultures on glycerine, agar, and broth. In some of their experiments they used bacilli that had been [illegible] from their soluble products, but they found that the results were the same whether they used bacilli from agar or broth, and whether these had been washed free from their soluble products or not. They concluded that the lesions produced were due to some substance—bacteria-protein—set free by disintegration of the microbes in the tissues, or extracted in some other way. They incline more to the former view.

By subcutaneous inoculation they produced an abscess in from two to six weeks. Tubercle bacilli stainable by the ordinary methods were found in the pus. By intraperitoneal and pleural inoculation with a milky emulsion of bacilli they produced nodules of various sizes on the serous membranes. These were made up of a central creamy-looking part surrounded by fibrous tissue. The central part consisted of epithelioid cells and giant cells. Tubercle bacilli were abundant in the central part; well-marked caseation was not found.

Intravenous inoculations were made into the auricular veins of rabbits. The animals were killed and examined at intervals of from one to sixty days. A few died after the third week. In animals killed after one day the bacilli were found in the lungs, liver, and spleen, most abundantly in the first-mentioned organ. The older the tubercle the fewer seemed to be the bacilli. After five days white nodules were found in the lung, some being microscopic, others quite visible to the eye. They were present up to the sixtieth day, the longest period of observation. Their structure consisted of epithelioid cells, giant cells, and leucocytes. Bacilli were found between the cells and inside giant cells. Later the nodule was denser and made up of epithelioid cells and loose connective tissue. After three weeks microscopic nodules, apparently having their origin inside the capillaries, were found in the liver. After five or six weeks visible nodules were found in the latter organ. Prudden and Hodenpyl conclude that the nodules originate in a proliferation of the vascular endothelium under the stimulus of dead and disintegrating tubercle bacilli. They say that "the dead bacilli seem to act as foreign bodies simply, curiously stimulating, it is true, but only foreign bodies after all." I think too much is made here of the foreign body question. Every foreign body does not produce this tissue reaction, although the power to do so may not belong exclusively to the tubercle bacillus. The action of the dead tubercle bacillus is in large part, at least, special to it, and it is the foreign body of this order that we are most likely to meet with in the tissues.

At the conclusion of their paper these authors offer some suggestions as to the influence of the products of the living germ on the degenerative changes in a true tubercle. They suggest, too, the possibility of the more fibrous tubercles being due to dead bacilli.

Curiously enough it was that last idea and a wish to further test an opinion which I had formed as to the origin of tuberculous giant cells, that led me to undertake some experiments with dead bacilli. I had conceived the idea and started experimenting with dead bacilli before I knew of their paper. I have since read it very carefully, and wish here to acknowledge my indebtedness to the authors. I have performed most of my experiments on different animals, but many of the results confirm those of Prudden and Hodenpyl.

I must also mention that Straus and Gamaleia[3] have to some extent confirmed the results of the American authors by experimenting on guinea-pigs, rabbits, and dogs. They say little about lesions in the liver. They did not find giant cells in the nodules. That may be because in their experiments too few bacilli were arrested in one part, or because the centres of the nodules were not examined. Many of their animals wasted and died. When the clumps of bacilli were broken up and well distributed in the fluid the animals wasted and died all the same, but no lesions were found. Evidently several dead bacilli are necessary in one part to produce a lesion. If the number of bacilli was very small the animals wasted, then recovered, and appeared quite healthy, but if a second small dose were administered they wasted and died. By using very small and ever-increasing doses of the dead bacillus they immunised the animal against it. With the products of the bacillus in artificial culture they could produce no lesion.

TUBERCULOSIS OF THE UDDER.

If one examines a number of tuberculous udders from the cow, one generally finds distinct caseous nodules, but that is not the only form that the tuberculous lesion may assume. At the Edinburgh abattoir during the last five years I have found a considerable number of very cirrhotic udders without any appearance of a caseous nodule in their substance. On examining these microscopically I have found tubercle bacilli and tuberculous giant cells, although the former were not very numerous. For that reason I think that the percentage of tuberculous udders has been slightly underestimated. These udders, however, will not very much increase the amount of tubercle-infected milk, because the affected quarters give little or no milk, and the cow is soon to be sent the abattoir. The bacilli enter the udder by way of the blood vessels, and it is highly probable that at an earlier date there were distinct caseous tubercles in these udders now cirrhotic. Two possible explanations of the difference in these two lesions suggest themselves:

1. The bacilli might have been overcome by the tissues to such an extent that they could no longer produce distinct caseation, but were still able to excite a proliferation of the tissues and cause the formation of giant cells.

2. The bacilli might have arrived in the gland in an attenuated condition, and were thus no longer able to produce the distinct caseous lesion.

There is nothing wildly imaginative in these suggestions if we think on what we know of the tubercle bacillus. It is well known that the bacillus first acts by exciting a proliferation of the tissues. The new cells, instead of completing their development, however, tend to become caseous. Still, we know that the cells do sometimes complete their development and form fibrous tissue. This is especially the case in the old tubercles found in the ox and pig. They are often surrounded by a rim of fibrous tissue, which is invading the caseous centre. Moreover, the tuberculous lesions of the muscles—muscle is considered a bad medium for the growth of the tubercle bacillus—which I have described in the pig were distinctly fibrous.[4]

I may say, too, that I have once found tubercle bacilli in fibrous nodules under the mucous membrane of the fourth stomach of the ox, and have several times found them in fibrous thickenings on the subcarpal regions of the ox. These are mainly the observations which have led me to undertake these experiments.

Experiment I.—Irish terrier bitch. I began to experiment on this animal for the purpose of trying to render her immune against tuberculosis by using the toxin and dead bacilli. I first injected her with tuberculin in doses varying from 10 c.cm. to 30 c.cm. There was no temperature reaction. I then continued with injections of the products of the tubercle bacillus in glycerine broth, from which the microbes had been separated by filtration through porcelain without previous heating. The cultures were allowed to go for from 2 to $2\frac{1}{2}$ months and longer. Beginning with 6 c.cm., I went to 10, 20, 40, 50, 95 c.cm. without disturbing the animal at all. With 95 c.cm. the temperature rose 0.8° F., but was not above the physiological limits (102.6). I next gave her 190 c.cm. into the vessels and under the skin. This toxin came from a $2\frac{1}{2}$ months' culture. It produced a rise in temperature of 1° F. in $6\frac{1}{2}$ hours. This shows the feeble systemic effect of the tubercle toxin. Thinking that the toxin prepared in the ordinary way was too weak to produce much effect, for there followed no local lesion whatever—it thus differs from the powerful diphtheria toxin—I thought of trying the effects of dead bacilli. Under the skin of the chest region I injected a thick emulsion of tubercle bacilli killed by three hours' steaming. The bacilli came from a glycerine broth culture that had been kept for for five months. An abscess formed and burst three days afterwards. A month later a second injection of bacilli in thick emulsion was given under the skin. The latter came from a glycerine broth culture that had been in the incubator for four months, and afterwards killed by three hours' steaming. On the third day a swelling about the size of a hen's egg formed at the seat of inoculation. The skin over this abscess was asepticised, and some pus was aspirated into a sterilised syringe by passing its needle into the cavity. Cover-glass preparations made with the pus simply swarmed with tubercle bacilli; no other microbes were present. Glycerine agar tubes sown with the pus remained sterile. The abscess then was caused by the tubercle bacilli, which were beyond doubt dead. The abscess burst on the fifth day, and the wounds took a considerable time to heal up. The temperature rose to 105° F. on the morning after the second injection. It fell to 101.5° on the sixth day. On the afternoon of the sixth day 30 c.cm. of a filtered tubercle culture were injected. The temperature next morning had risen to 104.5°, and had fallen to 102° the day after. Three weeks after the second injection of bacilli I injected subcutaneously about 4 c.cm. of the dregs left in the preparation of tuberculin. This material was very rich in bacilli, which had been killed at about 110° C. Five days afterwards there was slight swelling and tenderness, but no distinct abscess formed. The temperature rose 1° on the day after, and fell to the normal on the fourth day.

One might draw the following conclusions from this experiment: (1) That the soluble products of the tubercle bacillus produce little effect on the healthy organism, although they have a very decided action on animals whose bodies contain the tubercle bacillus, living or dead. (2) That the dead bacilli are far more active than the soluble products, although this may be on account of their retaining a strong toxin in their bodies.

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